

## Guidance for management of companion animals that have been exposed to a human with Ebola virus disease

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For the Canadian working group of Ebola virus in animals

### Introduction

*Zaire ebolavirus*, more commonly referred to as Ebola virus (EV), is one of five species of the Genus *Ebolavirus*, along with *Bundibugyo ebolavirus*, *Reston ebolavirus*, *Sudan ebolavirus* and *Tai Forest ebolavirus*.<sup>1</sup> Ebola virus resides is naturally found in central and sub-Saharan Africa in wildlife, with fruit bats of the Pteropodidae family believed to be the reservoir hosts.<sup>2-4</sup> Ebola virus disease (EVD), previously referred to as Ebola hemorrhagic fever, is a highly fatal infection in humans and some animal species, with reported mortality rates of 25-90% reported.

The 2014 EVD epidemic in West Africa, along with the presumed ongoing risk of emergence of this zoonotic virus from its wildlife reservoir in West and Central Africa have led to questions being raised about the potential for infection of domestic, wild and feral animals in Canada, with concern about infected animals to serve as a source of subsequent human infection. The most plausible mechanism of introduction of Ebola virus (EV) into Canada is via a person who was infected in an endemic region; yet, given the high incidence of animal ownership and other animal contacts in Canada, there is some potential for exposure of an animal to EV should an infected person enter the country.

Human health risks from animals exposed to EV are two-fold. One is the potential for animals to become infected and then transmit the virus. There are significant knowledge gaps pertaining to the host range of EV and whether domestic animals are susceptible to infection and are able to transfer EV to

humans or other animals (Appendix 1). In the absence of evidence to the contrary, the precautionary principle indicates that one should assume that a wide range of animal species are susceptible to EV infection and capable of transmitting the virus once infected. Yet, it should be remembered that there is currently no evidence indicating a risk from most domestic species, with the exceptions being pigs and non-human primates.<sup>5,6</sup> The other potential concern is for animals to be transiently contaminated with the virus from a symptomatic person (e.g. exposure to vomit) and act as a fomite. While EV survives for only short periods of time outside the host, the potential for fomite-based transmission of EV cannot be dismissed.

For the purposes of this document, ‘companion animal’ refers to any animal kept in a household, including exotic animals and pet pigs (e.g. Vietnamese pot-bellied pigs). However, quarantine of some species (e.g. non-human primates) may be impractical in many situations because of the handling and management requirements and heightened risk because of the known susceptibility to EVD. Significant case-by-case decision-making is required because of the potentially significant differences between species, between individuals of the same species, between facilities and in the ability to establish a proper quarantine facility. If assurance of the ability to safely and effectively quarantine an animal cannot be made, euthanasia would be indicated.

### **Identification of Animal Contacts of People with Known or Suspected EVD**

It is important that animal contact (not just animal ownership) since onset of symptoms be queried of any individual with EVD. Public health personnel involved in contact tracing of persons under investigation (PUI) for EVD, as well as probable or confirmed EVD should query any animal contact (inside or outside the household) that occurred after the onset of symptoms that could be consistent with EVD. If any animal contacts are identified, discussion should occur between the relevant public health and animal health personnel (which will vary by province/territory) to determine whether there has been potential exposure of animals. Persons making the initial identification of potential animal exposure should recommend that animals be kept in the house or on the farm until relevant animal and public health personnel have investigated and determined whether other measures must be taken.

### **Identification of Exposure**

There is no standard definition of what constitutes exposure of an animal. As with human-human transmission, it is assumed that the risk of exposure starts once the infected person develops symptoms of EVD, with lesser risk in the first few days of infection compared to when more advanced disease is present. Simply living in the same residence with a person with EVD symptoms does not necessarily indicate that an animal was exposed; however, given the regular and undocumented nature of human-pet contact in households, unless it is certain that the infected individual did not have direct contact with the animal and that there was no indirect contact of the animal with potentially contaminated surfaces, the animal should be assumed to have been exposed. Close contact with a human confirmed Ebola patient since the onset of the patient’s symptoms, including sitting in a lap, being cuddled, being kissed, licking, sleeping in physical contact or contact with mucous membranes presumably constitutes a higher risk of exposure. Questions should be asked for the time period since the confirmed Ebola patient’s symptoms began. Exposure to blood or body fluids (e.g. vomit) of an Ebola patient would constitute a particularly high risk of exposure.

## Confinement facility requirements:

An adequate containment facility must be identified in advance.

- A quarantine facility must provide at least two (and ideally more) physical containment levels (e.g. crate/kennel within a secured building). The facility should be locked at all times.
- Specific containment zones should be defined and physically delineated (e.g. taping floors). These consist of:
  - *The clean zone*: A controlled area that contains the support functions necessary to the activities on the quarantined premises. This area is not considered contaminated with the EV. Personal protective equipment (PPE) is not required in this zone.
  - *The buffer zone*: The area between the clean and hot zones, where contamination reduction takes place. This would include the area where PPE doffing occurs and any area transited with the animal or by personnel in potentially contaminated PPE (prior to disinfection). It is important to remember that the buffer zone must be considered contaminated, albeit at a lower level than the hot zone. Clear boundaries should be used to show the limits of the zones. Physically indicating the barriers (e.g. taping off the area) may be useful.
  - *The hot zone*: This is the area that may have been contaminated with EV from the potentially infected animal. This would consist of the containment room, plus any areas used to store waste or other potentially contaminated items. To enter this zone, a person must wear full PPE and upon exit, this person is considered contaminated, regardless of what was done or touched inside the zone.
- The animal must be kept indoors at all times.
- Facilities with pest infestation must not be used.
- Specific quarantine housing measures will be dictated by the size and species. However, there must be adequate space for the animal to eat, drink, urinate, defecate and move around, and for it to be properly managed.
- Specific means of animal handling and management will vary depending on the facility and animal. The goals are to provide an acceptable quality of life while maintaining biocontainment and facilitating management. In some situations, the animal may have to be kept in its crate/cage for urination and defecation. For dogs, when possible, providing a contained area outside the cage/crate (e.g. partitioned area within the quarantine room) or one crate for living and one for urinating/defecating, should be considered to help keep the animal's primary living space clean and to improve the animal's quality of life. Whenever possible the animal should be provided the same diet as it was previously eating to reduce the likelihood of gastrointestinal abnormalities that might be confused with signs of EVD.

## Preparation phase

All steps in the process of retrieving an animal and establishing quarantine must pre-planned, with clear roles and responsibilities for all personnel that are involved. All required supplies must be available, along with adequately trained personnel.

## Identification and training of caretakers

- Animal care should be provided by as few individuals as possible, with a minimum of two being present every time the containment area is entered. Unless two people are required for an activity, one person should stay outside of the hot zone to observe the other's actions and be able to intervene in the event of a problem (e.g. suit breach, bite). Depending on the animal's temperament, the procedures being performed, the nature of animal housing (e.g. fully contained in a cage or being taken outside of the cage or otherwise handled) and the facility, the observer might require full PPE, be able to observe in the buffer zone while wearing partial PPE (e.g. boots and gloves) or be able to observe from the clean zone without PPE.
- Caretakers must have experience with animal handling (appropriate for the species) and be trained on proper use of PPE (including donning and doffing).
- Auditing of PPE donning and doffing practices should be performed prior to animal quarantine and periodically during the quarantine period.

## Personal protective equipment (PPE)

PPE shall consist of, **at a minimum**

- Double gloves, with outer glove taped to the suit with moisture-resistant tape
- A splash resistant disposable hooded suit, with foot covers impervious to fluids either as part of the suit or attached to the suit with water-proof tape
- Eye protection (goggles or face shield)
- Air purifying respirator (N-95 or equivalent level of protection)
- Additional protective equipment might be required in certain situations (e.g. heavy duty or puncture-resistant outer gloves, head or neck protection if not sufficiently provided by the protective suit)

## **PPE training and use**

- Personnel should be trained in the proper use of PPE, including mask fit-testing.
- Use of structured checklists should be considered, particularly during doffing.
- PPE should be donned and doffed in an appropriate manner (Appendices 4 and 5).
- An alcohol-based hand sanitizer should be applied after each PPE removal step

## **Information to be collected prior to animal contact**

Prior to contact with the animal(s), information about the animal(s) should be obtained to determine whether quarantine will be feasible and to facilitate recovery and transportation of the animal, and subsequent quarantine. Information that should be obtained initially includes:

- Species
- Breed
- Sex
- Age
- Colour/markings
- Whether a microchip is present
- Health status, including vaccination history, any required medications and contact information for the animal's veterinarian
- Temperament
- Diet (including specific brand and product names)
- Contact information for an alternate decision-maker in the event that the owner is unable to make decisions. Formal (written) designation of someone to act as official agent should be considered so that there are clear decision-making plans in the event that discussions of euthanasia or other major decisions must be had.
- Areas the animal has visited and people or animals the animal has had contact with since the onset of the person's symptoms and the timing of these events in relation to the date of onset of the person's symptoms. Any potential contact with humans outside the household should be forwarded to public health personnel.

Once this information has been collected, consultation between the relevant public health and animal health personnel will be performed to determine whether the animal must be quarantined and to identify any specific issues or concerns that might impact the ability to safely and effectively quarantine the animal(s). In some situations, quarantine might pose a significant risk to the pet's health or welfare. This would include animals that behaviourally will not tolerate confinement, those whose temperament would make containment very difficult (e.g. aggressive), animals that require medication which cannot be given remotely (e.g. injectable medications, medications that must be given directly per os and not in food, water or treats) and species where management in a quarantine facility might be impossible (e.g. some exotic species, non-human primates). Assessment of the medical and welfare risks of quarantine will be made by the animal health and public health personnel and the owner/agent, in consultation with any relevant veterinary experts.

As part of the initial investigation, whenever possible, the owner or someone else in the household should be instructed to confine the animal to a cage, crate or small room (e.g. bathroom) to provide an

extra level of containment on order to prevent escape from the household and to facilitate retrieval of the animal.

### **Retrieval Planning and Supplies**

Retrieval and transportation procedures and timing should be coordinated with the quarantine facility. Retrieval procedures should be planned in advance, considering the number of animals, species and any available information about size and temperament. An inventory and procedure checklist should be completed prior to departure to the animal's location (Appendix 2).

## **Implementation phase**

### **Pre-departure procedures**

- Provide a vehicle with a secure compartment of a vehicle that is separate from the driver compartment (e.g. locked trailer, moving van, covered pick-up truck bed).
- Ensure that there is no need to stop (e.g. for fuel) unless absolutely required (e.g. trip requiring more than one tank of fuel). If a stop is required, the vehicle must not be left un-attended at any time.
- If there might be a need for crowd control, the appropriate police services should be contacted prior to departure to the animal's location so that they can be present at the time of arrival.

### **Establishment of Biocontainment Zones at the Animal's Place of Residence**

Containment zones should be established as early as possible. Initial planning of containment zones can be made prior to departure based on information obtained from public health personnel and the animal's caretaker. Upon arrival, these should be reviewed and revised as needed prior to leaving the vicinity of the vehicle. The following zones should be established, as is discussed in more detail above: clean zone, buffer zone and hot zone.

The clean zone is the preparation area that is not potentially contaminated by the animal or personnel that have been in the hot zone. PPE is not required in this zone.

The buffer zone would include the area from the residence to the vehicle that is transited with the crated or caged animal. It is important to remember that the buffer zone must be considered to be contaminated, albeit at a lower level than the hot zone. The hot zone is the area that may have been contaminated with EV from the infected person or the animal. Typically, this would consist of the entire residence. Occasionally, additional areas (e.g. vehicle, porch, yard) might be considered contaminated. Public health personnel would obtain information about potentially contaminated areas during the course of initial investigation. To enter this zone, a person must wear full PPE and upon exit, this person is considered contaminated, regardless of what was done or touched inside the zone.

Clear boundaries should be used to show the limits of the zones. Physically indicating the barriers (e.g. taping off the area) may be useful, particularly in areas where bystanders may be present.

### **Retrieval and transportation protocols**

Personnel involved with retrieval of the animal must be wearing personal protective equipment (PPE), as described below, and have been properly trained in PPE practices.

- The vehicle should be parked as close as possible to the door of the residence, while still providing sufficient space for creating clear clean and buffer zones.
- PPE must be worn when entering the residence.
- Care and patience must be used to reduce the risk of PPE breaches from aggressive or excited animals.
- The door to the residence should be opened with caution in case the animal is loose.
- Care and patience must be used when retrieving the animal if it is free within the household or in a room. The animal may be disturbed by the presence of strangers and the appearance of PPE, and may be less predictable than normal. If the animal appears aggressive, fearful or there is a concern about any situation where catching the animal might cause a PPE breach,

alternative methods (e.g. pole snare, lure animal into crate/cage with food, live trap) must be considered.

- Once the animal has been located in the residence, it should either be:
  - Placed in a crate or carrier that is then secured with a lock. The crate or carrier must be inspected to ensure there are no defects that could result in escape.
  - Restrained by a leash attached to a securely-placed collar that cannot be removed by the animal during struggling and placed in a cage or crate located immediately outside the door of the residence. *This should only be done in situations where the size of the animal and crate is such that it cannot be carried through the door and to the vehicle.* If the animal must be walked outside to get to the crate (e.g. large dog in an apartment building with no elevators and where the crate cannot be carried down the stairs).
  - Consideration should be given to having two leashes on separate collars.
- The crate should be covered with an impermeable barrier to reduce the risk of aerosolization of EV that might have been deposited on the animal's haircoat (e.g. if the animal shakes or jumps around) and to guard against the very unlikely event that an infected companion animal is able to aerosolize the virus during activities such as barking or hissing, or in feces or urine that might be splashed during transportation. If possible, an absorbent pad should be placed in the bottom of the crate to absorb any body fluids.
- No other items such as beds or toys should accompany the animal.
- If a leash is used, it should be left in the house or placed in a biohazard bag and transported to the facility in a sealed container.
- The animal should be transported in a secure compartment of a vehicle that is separate from the driver and passenger compartment (e.g. locked trailer, moving van).
- After the animal has been placed in the vehicle, the buffer zone should be sprayed with disinfectant (Appendix 3) by an individual wearing PPE. If urine, feces or any other body fluid contaminate the buffer zone, the contaminated area should be soaked in an appropriate disinfectant for the indicated contact time, as much material and liquid as possible should be removed and placed into a biohazard bag, and the area disinfected once more. PPE must be worn during this process.
- PPE should be removed and hand hygiene performed before personnel enter the vehicle.
- Care should be taken to ensure that there is no need to stop (e.g. for fuel) unless absolutely required (e.g. trip requiring more than one tank of fuel). If a stop is required, the vehicle must not be left un-attended.
- After the animal has been transferred to the quarantine facility, the trailer should be cleaned and disinfected with an appropriate disinfectant that is known to inactivate EV or, alternatively, has a label claim of efficacy against non-enveloped viruses (Note: While EV is enveloped, non-enveloped viruses are more hardy so using a product with a claim against non-enveloped viruses will provide an extra level of assurance (Appendix 3)).



## Containment phase

### Quarantine period

- While no information is available for companion animals, a minimum 21 day quarantine is recommended based on data from humans. This period starts from the last potential direct or indirect contact with a person with EVD or potentially EV-contaminated environment (normally at the time of removal from the household).
- This period may be extended if further information about EVD in animals is obtained or based on the clinical status of the animal, such as if signs that could be suggestive of EVD (e.g. non-specific systemic illness, gastrointestinal signs, respiratory disease, hemorrhage) are identified late in the quarantine process.

### Handling Procedures

- Entry into the confinement area and contact with the animal should be minimized.
- For each animal, a plan should be developed for any handling and other management steps to reduce the risk of contamination of the environment and personnel. Specific feeding, cleaning and other management practices will vary with the animal species, size temperament and the facility.
- Aerosol-generating procedures must be avoided (e.g. hosing).
- Any bodily wastes should be removed without direct contact (e.g. shovel, disposable absorbent pads, mop) and an appropriate disinfectant should be applied to any potentially contaminated surfaces.
- No items (e.g. disinfectant bottles) apart from PPE, properly managed garbage (see below) and diagnostic specimens may leave the hot zone.

### Caretakers

- PPE must be worn whenever the hot zone is entered, regardless of what procedure or contact is planned..
- Caretakers must self-monitor for fever (temp  $\geq 38^{\circ}\text{C}$ ) and report any fever or other clinical abnormalities that could be consistent with EVD to public health within 12 hours.
- A log of caretakers should be kept, including dates that they cared for the animal.
- Any adverse events (e.g. PPE breaches, bites, scratches) that could result in potential EV exposure must be reported to public health within 24 hours.
- The caretaker team will provide regular updates to the owner or their designated agent. Pictures may be taken provided that they can be taken from outside the hot zone or with a device that does not leave the room (e.g. mounted closed circuit camera) or that is used in a sealed covering that allows for immersion in disinfectant as part of the exit process.

### Health monitoring of the animal

- The animal's attitude, appetite, behaviour and other external findings should be recorded at least twice daily. Direct examination of the animal should be limited to situations where it is absolutely required.
- A veterinarian must be designated for the oversight of the animal's care and confinement, if one is not already part of the caretaker team.
  - If the veterinarian must have any contact with the animal or enter the hot zone, the same requirements for PPE use and training apply.
- If clinical abnormalities are noted, the caretaker team, veterinary personnel and federal/provincial/territorial animal and public health personnel that are involved in the quarantine will determine a plan of action.
  - This will typically involve taking of the animal's body temperature. This should be done in pre-determined manner to minimize the risk of bites, scratches or aerosolization of material from the animal's haircoat. This may involve chemical sedation. Regardless of whether or not sedation is used, a muzzle should be applied.
  - If there is any suspicion that the clinical abnormalities could relate to EV infection, Ebola virus testing is indicated. This will be performed by the National Centre for Foreign Animal Disease. Preparations for sample shipment and testing must be confirmed before sample collection.
  - Samples must be collected for diagnostic testing, as described below. At a minimum, chemical sedation should be used to facilitate sample collection, ideally administered in food or treats. Remotely administered (e.g. pole syringe) heavy sedation or general anesthesia may be appropriate.
  - Even if EVD is not considered to be a potential cause of the clinical abnormality, collection and submission of specimens for other diagnostic testing will not be performed because handling requirement for specimens would preclude testing in a veterinary diagnostic laboratory.
  - The caretaker team will determine any treatment regimen for an animal displaying clinical abnormalities. Minimal treatment will be provided; in particular, treatment that requires close contact with the animal and/or use of sharps should be avoided. This should be decided on a case-by-case situation, considering the type of disease, desired treatments, risk that the animal was exposed to EV and availability of adequately trained personnel.

### **Waste disposal**

- The primary containment enclosure (cage, crate) will typically be cleaned a minimum of once daily; however, in some circumstances, less frequent cleaning may be reasonable (e.g. cat with litterbox that is not very soiled)
- All materials that are to be disposed of, including leftover food, body waste and consumable items, must be placed in a biohazard bag within a leak-proof container (e.g. plastic bin, garbage can).

- Transportation of waste outside of the facility must be coordinated, including securing any permits that may be required. At a minimum, biohazard bags will be sealed, double bagged and placed inside a leakproof container that is sprayed with disinfectant as part of the exit process.
- If space permits, waste should be stored in the hot zone until the quarantine period is complete, and then all of the waste can be removed at once.

## Testing

Little is known about optimal methods for testing companion animals. Testing of blood is presumably the most sensitive approach but should be limited because of the risks of exposure of personnel during sampling and sharps handling. A sedation or anesthesia plan must be made before collection of samples that require direct contact with the patient. In addition to sedation/anesthesia, additional levels of physical restraint (e.g. muzzle, cat bag) should be used to reduce the risk of bites or scratches, or needlestick injury from animal movement.

The Canadian Food Inspection Agency (CFIA), National Centre for Foreign Animal Disease (NCFAD), is responsible for sample testing in Canada. See the CFIA document “Procedure for the Submission of Ebola Virus or suspect Ebola Virus Diagnostic Samples from Animals” for more information regarding sample submission. Samples should be collected into plastic (not glass) containers or tubes. The testing recommendations are as follows:

### *Routine testing during quarantine*

- Urine and feces/rectal swabs on or about Day 7. If possible, depending on the animal’s temperament and handling facilities, an oral swab can also be collected.
- Oral swab, nasal swab, feces/rectal swabs, urine (if possible), serum and whole blood on or about Day 18 (to enable release on day 21 with negative results).
- Rectal swabs are preferred over feces but because of the increased contact that is required for rectal swab collection, decision of which to collect should be done on a case-by-case basis, depending on the animal’s temperament and the ability to safely collect a rectal swab.

### *Testing in response to the animal developing signs potentially associated with EVD*

- Serum and whole blood, along with oral, nasal and rectal swabs should be collected.
- If initial results are negative but clinical abnormalities potentially attributable to EVD persist, consideration should be given to repeating sampling greater than 72h after the onset of clinical signs.

### *Death or euthanasia of an animal during quarantine*

- Regardless of the cause of death or reason for euthanasia, animals that die or are euthanized during quarantine will be considered potentially infected with EV.
- Rectal swabs, serum, whole blood, nasal and oral swabs (and urine, if possible) should be collected *post mortem* and submitted for EV testing.
- Submitted tissues from swine (including pot-bellied pigs) should include lung, trachea and associated tissues. For species other than swine, additional tissues may also be submitted, since tissue distribution is unknown in other species. However, there should be consideration of the likelihood of EVD, the value of the added samples and the ability to safely conduct a necropsy when determining whether to test additional sample types.

- Consideration can be given to submission of the body to NCFAD for more comprehensive testing only under very special circumstances. This must be based on prior discussion and agreement with NCFAD to determine if testing is considered possible and useful, and if shipping requirements can be met.
- The body should be incinerated.

### *Diagnostic sample handling*

After collection and while still in the hot zone, the sample collection containers should be sprayed with disinfectant solution and then placed in a receptacle (bag or container) in the buffer zone and sealed. The outside of this container should then be wiped or sprayed with disinfectant, and then the container should be placed directly into a durable, leak-proof container that is being held in the clean zone. Samples should be prepared for shipping according to Transportation of Dangerous Goods Regulations using a triple packaging system. This consists of placing the sample container inside a sealable specimen bag (primary container), which is wrapped with absorbent material and placed in a water-tight, leak-proof secondary container, then placed in an outer shipping package.

### *Shipping*

Samples should be packaged and shipped in consultation with NCFAD and the chosen courier company. The company must be Transportation of Dangerous Goods (TDG) certified.

### *Additional testing considerations*

Testing approaches may vary between animals (e.g. species, age, temperament) and where feasible, collection of additional samples for PCR or other testing (e.g. serology) could be considered to obtain more information about natural infection (or lack thereof) in companion animals. Collection of blood samples must be performed with utmost caution and would typically involve, at a minimum, chemical restraint. Remotely administered general anesthesia may be appropriate in some circumstances. It must be recognized that practices not typically considered acceptable in a veterinary hospital (e.g. use of certain injectable agents) may be appropriate in handling an animal being quarantined because of the risk of exposure to a pathogen of significant public health consequences.

## **Communications**

A communication plan should be established as early as possible, through discussions amongst the owner/agent, caretakers and federal/provincial/territorial animal and public health personnel. This involves communication between team members, communication with the owner/agent and communication with media and other outside bodies.

## **Minimum Criteria to Release Pet from Confinement**

Animals will be released from quarantine after the following have been fulfilled:

1. A minimum of 21 days from the last potential exposure to an EVD-affected person or a potentially EV-contaminated environment.
2. Negative EV test results (specific tests determined by NCFAD) from Day 18-21.
3. No clinical signs potentially attributable to EVD are present.

If an animal tests positive for EVD, discussions must be held between the caretakers, veterinary personnel and federal/provincial/territorial animal and public health personnel to determine the appropriate course of action. This may involve euthanasia or continued monitoring to obtain evidence about EV shedding in the particular species, depending on the severity of disease, animal welfare and willingness of caretakers to continue handling a known EV-infected animal. . It should be recognized that there is limited information on the validity of some of these tests in various species, and that false positive results are possible. If continued management of the animal is chosen, determination of how (or if) to declare an animal EV-free and eligible for release from quarantine will be made by public health and animal health personnel, veterinary caretakers and any relevant experts, as a single negative test may not provide the acceptable level of assurance in a situation when an animal is known to have been infected. Determination of how (or if) to declare an animal EV-free and eligible for release from quarantine will be made by public health and animal health personnel, veterinary caretakers and any relevant experts.

### **Terminal cleaning and disinfection**

- At the end of the confinement period all linens, pet beds, and other textiles used in the confinement facility should be discarded as biohazardous waste, even if the animal was released uninfected, because of the inability to guarantee there was no transient EV shedding. Because of the poor environmental survival of EV and lack of evidence of subclinical shedding by species other than reservoir hosts, this is done out of an abundance of caution, with an understanding that there is limited true risk.
- Items that will not be discarded (e.g. cages, disinfectant sprayers) will be thoroughly cleaned and disinfected by individuals wearing PPE. Ideally, more than one round of disinfection will be performed, and the items and environment will be left unused for 7-14 days. This is also done out of an abundance of caution and may be excessive, but when practical, it can provide more assurance to individuals who will subsequently be in contact with the equipment or area.

## Appendix 1: Table of information pertaining to Ebola virus infection in animals

Species	Natural infection	Incubation period	Clinical signs	Duration of infectivity	Comments	Ref
Fruit bats (various species)	Yes	Unknown	None reported. Probably none as true reservoirs.	Unknown. Transitory viremia noted in experimental study of some species.	Thought to be reservoir hosts. Unknown how virus bridges to other species.	2-4,7
Humans	Yes	2-21d typically reported. Mean of 11.5-12.7 d estimated from outbreaks, with indication that 4-5% of people might have incubation >21d.	Initial non-specific signs such as fever, fatigue, diarrhea, vomiting, rash; progressing to coagulopathy, shock, electrolyte imbalances and other systemic signs	Onset at the time of first clinical signs. Peak infectivity after initial non-specific signs.	Virus shed in blood, saliva, vomit, feces, breast milk, tears and semen. Typically 40-90% mortality.	8-11
Non-human primates <sup>a</sup>	Yes	7-12 days	Fever, classical hemorrhagic fever signs	Presumably similar to humans	High mortality rates with decimation of some family groups. Experimentally, mortality rates of up to 100% with some animal species and virus species (e.g. <i>Zaire ebolavirus</i> and macaques)	6,12, 13
Pigs <sup>b</sup>	Unknown	4-7d	Fever, tachypnea, anorexia, lethargy	Unknown. Virus detected in blood, nasal, oral and rectal swabs. Shedding from the oronasal mucosa continued for 2 weeks. Possible aerosol transmission.	Concern expressed about aerosol or airborne transmission in experimental situations because infection often localized to the respiratory tract. One study strongly suggested aerosol transmission from piglets to macaques.	5,6
Guinea pigs	Unknown	Lab adapted virus, ~2d	Variable with wild virus; often transient febrile illness. With lab adapted virus, disease similar to that in humans (without rash) can be produced.	With lab adapted virus, starts on day 2 and guinea pigs typically die by days 7-9 post-infection so extend of shedding not well understood.		14-16
Laboratory mice	NA				Quite resistant to infection by wild virus. Laboratory adaptation required to cause disease.	16-18

Duikers	Yes (maybe)	Unknown	Unknown	Unknown	Ebola virus identified by PCR in one dead Duiker as part of wildlife surveillance around a human outbreak in the Republic of Congo. Seems to be based on single dead animal with same case published twice. More anecdotal evidence relates to commonness of dead duikers in conjunction with NHP deaths in areas with active Ebola virus disease (EVD).	12,13,19
Porcupines	Maybe	Unknown	Unknown	Unknown	WHO fact sheet lists contact with porcupines as a potential source of human infection, but I am unaware of any published supporting data. May relate to observations of many dead porcupines with disease suggestive of EVD in areas with human outbreaks and concurrent wildlife deaths.	20
Equids	Unknown	Unknown	Unknown	Unknown	3/13 donkeys seropositive in Central African Republic	21
Dogs	Maybe	Unknown	Unknown	Unknown	Only evidence of infection is seroconversion, with high rates reported in villages in Gabon with human outbreaks and concurrent animal deaths. No recovery of virus or viral antigen.	22

Wild rodents	Maybe	Unknown	Unknown	Unknown	Partial Ebola virus sequences recovered from mice and a shrew. Negative results obtained with sampling large numbers in D.R.Congo.	23
Arthropods	Unknown	Unknown	Unknown	Unknown	Unsuccessful attempts at experimental infection; however, Marburg virus can persist in <i>Aedes</i> mosquitoes for > 3 weeks after experimental infection.	7,19, 24

<sup>a</sup> Includes gorillas, chimpanzees, macaques, mandrills, baboons

<sup>b</sup> *Zaire ebolavirus*, not the related but separate species *Reston ebolavirus*



## Appendix 2: Suggested inventory list for retrieval of an animal from a household and transportation to a quarantine facility.

### PREDEPARTURE CHECKLIST

Animals to be picked up		
	Number	
	Species	
	Size/weight	
	Location	
	Demeanor (for retrieval by strangers, transport in truck)	
	Sedation required (dose)?	
	Diet (must be obtained)	
	Additional medical history/ concerns	
Notify local police		
	Crowd control needed at site?	
	Emphasize NOT to notify media until after	
Access to residence		
	Key vs code	
	Stairs vs elevator	
Gas in vehicle		
	If long distance, refill tank just prior to arrival at destination	
Personnel selection		
	Experienced handling species in question	
	Appropriately trained in PPE	
Other considerations		
	Arrangements for autoclaving garbage	
	Verify how cage will be secured in vehicle (ropes, bungees)	


**INVENTORY AT RETRIEVAL SITE**

✓	Item		Quantity	Notes
	Driver / crowd control		1	-This person is “clean” -Able to manoeuvre vehicle as needed
	Animal handlers		2	-Experienced with species involved, trained in PPE
	Sedation for animal, if necessary		Per weight	
	IF	Meatball/pill pocket (atravet)	1	
	OR	Pole syringe	1	
	Transport vehicle		1	-Separate compartment for transporting animal (e.g. covered pick-up) -Transport compartment should be clean and empty
	Heater (and power source) for transport compartment		1	Depending on weather
	Large tarps		As needed	If needed to provide shelter from wind/precipitation
	Cage for animal		1	-Appropriate size for handling/transport (must fit easily in transport compartment) -May be used subsequently in hot zone
	Cage cover		2	-Should cover all openings of cage to prevent aerosol contamination -Suggest surgical table drape (light, breathable, impermeable), bring one spare
	Bungee cords or rope		As needed	-For securing the cage in the transportation compartment
	Catch pole		1	In case needed
	Cat bag and/or cat mits		1	In case needed
	Muzzles (at least two sizes)		2	In case needed
	Leashes and collars		2 each	In case animal cannot be safely moved in cage to vehicle
	Rubber gloves (e.g. disposable dishwashing gloves)		2 pair	To protect thinner inner and outer gloves if moving a large animal crate
	PPE (6 sets total, 3 per person)			
		Tyvek coveralls	6	Sized to retrieval team members (3 each)
		N95 masks (disposable)	6	Sized to retrieval team members (3 each)
	OR	Silicone half-facepiece masks AND bag/boot/bin for disinfecting	6 1-6	-Sized to retrieval team members (3 each) -Masks will need to be bagged/binned after each use for disinfection upon return to base
	AND	N95 filters (disposable)	12	2 per person per use (up to 3)
		Face shields	6	1 per person per use (up to 3)
		Disposable shoe covers	18	2 per person per use (up to 3), plus extras for “clean” person if needed

	Disposable gloves – long cuff	24	-4 per person per use (up to 3) -Inner gloves can be short-cuff or long-cuff
	Duct tape roll	2	Consider extreme-temperature duct tape in cold weather
	Alcohol based hand sanitizer (pump)	2	1 spare in case bottle gets contaminated
	AHP spray bottle	2	Including full reservoir of disinfectant, 1 spare in case bottle gets contaminated
	AHP “pumper”	1	Including full reservoir of disinfectant
	Container for footbath	1	With AHP
	Large biohazard bag in plastic bin/can	1	Outside considered clean, inside considered contaminated once used
	Extra biohazard bags	2	In case needed
	Absorbent pads	12	In case needed (i.e. for clean up of urine or feces)
	Roll of yellow “Do not cross” tape	2	In case needed
	Seat for doffing	1	-Small bench or stool (no back), easily disinfected

Spares and “in case” items can be kept in the “clean” part of the vehicle until needed, and may be retrieved by the driver/crowd control person, who must therefore be familiar with the equipment and where it is kept.

#### INVENTORY AT QUARANTINE SITE – COLD ZONE

✓	Item	Quantity	Notes
	Animal food	21 day supply	-Less what is kept in hot/warm zones, if any -Feed diet as specified by owner to avoid diet change whenever possible, HOWEVER for logistical reasons it must be a readily-available commercial diet (canned or dry) which will be purchased separately (i.e. home-cooked/raw diets will not be used, food will not be brought from the residence)
	Can opener	1	If required for feeding canned food
	Food bowl for transfer	1	“Air drop” food into bowl in warm zone
	OR Disposable plates/bowls	42	-1 per feeding, twice daily for 21 days -Can only be used if animal will not attempt to eat the plate/bowl, or if the animal eats all the food at once and the plate/bowl is removed before personnel leave the hot zone
	Transfer water jug, 4 L	1	“Air drop” water into jug in warm zone
	PPE		<b>Counts below are MINIMUM required</b> , does not account for additional entries or procedures requiring additional precautions or personnel
	Tyvek coveralls	84	-2 persons twice daily for 21 days -Sized for 4 main quarantine personnel
	N95 masks (disposable)	84	-2 persons twice daily for 21 days -Sized for 4 main quarantine personnel

	OR	Silicone half-facepiece masks	4-8	-2 per person (1 in use, 1 being disinfected/dried) -Can be used by multiple persons if they wear the same size (disinfected after each use regardless), but consider sizes needed for each team of 2
	AND	N95 filters (disposable)	168	2 per person, 2 persons twice daily for 21 days
		Face shield (disposable)	84	2 persons twice daily for 21 days
		Disposable shoe covers	168	2 per person, 2 persons twice daily for 21 days
		Disposable gloves – long cuff	336	-4 per person, 2 persons twice daily for 21 days -Inner gloves (half) can be short-cuff or long-cuff
		Duct tape rolls	10	Estimate 3 rolls per week (for PPE and miscellaneous)
		Transfer disinfectant jug, 4 L	1	“Air drop” disinfectant into jug in warm zone
		Alcohol based hand sanitizer (pump)	1	Exit from doffing area
		AHP spray bottle	1	Including full reservoir of disinfectant
		Large biohazard bags	15	-Ideally minimize the number of separate bags used over the course of the quarantine, but also critical to avoid overfilling the bags as this could impede proper closure -Estimate maximum one bag per three days for doffing (warm one) and disposal of food/excrement (hot zone)
		Absorbent pads	60	To restock warm and hot zones (~2.5 per day, for urine, feces, miscellaneous spills)

Each member of the quarantine team must wear scrubs (top tucked into pants) and sturdy closed-toes shoes when donning PPE. After doffing, shoes should be dipped (soles) and sprayed with disinfectant (tops) prior to leaving the main cold zone to change clothes. Scrubs should be laundered after each entry, and personnel should shower before leaving the building.

#### INVENTORY AT QUARANTINE SITE – WARM ZONE

✓	Item	Quantity	Notes
	Transfer food bowl (if not using disposable)	1	“Air drop” food into bowl in warm zone
	Transfer water jug, 4 L	1	“Air drop” water into jug in warm zone
	Transfer disinfectant jug, 4L	1	“Air drop” disinfectant into pumper reservoir in hot zone
	Alcohol based hand sanitizer (pump)	1	Doffing area
	AHP spray bottle	1	Including full reservoir of disinfectant
	AHP “pumper”	1	Including full reservoir of disinfectant
	Plastic bin for disinfecting reusable face masks	1	If using reusable masks

	Large biohazard bag in plastic bin/ can	1	Full bags should be passed into the hot zone for storage until the end of the quarantine so all garbage can be autoclaved together (assuming this is manageable given volume of garbage and storage space)
	Container for footbath	1	With AHP
	Absorbent pads	2	
	Seat for doffing	1	-Small bench or stool (no back), easily disinfected -Personnel should doff one at a time in order to assist each other, therefore only one seat required

**INVENTORY AT QUARANTINE SITE – HOT ZONE**

✓	Item	Quantity	Notes
*	Cage for animal	2	-1 in use, 1 being cleaned/dried -If two cages of sufficient size are not available, could use transportation crate to temporarily contain animal while primary cage is cleaned and reset
	Soft rubber toys	1-2	Toys provided for environmental enrichment must be tailored to the individual animal, but no toys that could result in cuts/scrapes/bleeding (e.g. rope toys) or that the animal may destroy and ingest. Also suggest avoiding plush/absorbent toys if possible.
	Food bowl (if not using disposable)	2	1 in cage with animal, 1 being cleaned/dried and used for transfer of food from warm zone
	Water bowl	2	-1 in cage with animal, 1 being cleaned/dried -Consider hanging on cage door if will help prevent spillage
	Water jug, 4 L	1	Consider keeping full 21-day supply in room if feasible, but a 4 L (or smaller) jug should be available to facilitate filling water bowls
	Kitty litter	40 lbs	Consider using kitty litter as desiccant for garbage (to help reduce smell) even if the animal under quarantine is not a cat
	Bin for clean kitty litter	1	
	Scoop for clean kitty litter	1	
	Litter boxes	2	-1 in cage with animal, 1 being cleaned/dried -To minimize contact with urine/feces, litterbox should be filled with a minimal amount of litter, and when cleaned entire contents should be disposed and replaced with fresh litter in clean box, rather than “scooping” and leaving dirty litter in the box
	Towels for bedding	6	-To be disposed at the end of the quarantine or sooner if they become soiled -Can consider dog/cat bed after a few days if the animal is not a high risk for soiling or chewing on the bed and if it would improve the animal’s comfort level, but the bed would be disposed as for the towels
*	Alcohol based hand sanitizer (pump)	2	1 by exit, 1 by animal
*	AHP spray bottle	1	Including full reservoir of disinfectant
*	AHP “pumper”	1	Including full reservoir of disinfectant
*	Container for footbath	1	With AHP
*	Large biohazard bag in plastic bin/can	1	Full bags should be stored in the hot zone until the end of the quarantine so all garbage can be autoclaved together (assuming this is manageable given volume of garbage and storage space)

*	Absorbent pads	6	1 for under water bowl (assuming animal will not chew/eat the pad), 1 for removing gross urine and feces (from cage or run area) 4 spare at all times
	Extra pads and newspaper	2 & 2	To cover floor of “run” if enclosure set up for urination/defecation by animal (see note below)

\*These supplies can be the same as those used during the retrieval of the animal, after which they can remain in the hot zone for further use.

- Strictly limit inventory within the warm and hot zones, as everything in these zones will need to be autoclaved (including garbage) or terminally disinfected at the end of the quarantine
- For dogs, consider setting up a separate enclosed corner of the room into which the dog can be placed temporarily while personnel are in the room and cage is being cleaned/reset, and the floor of which can be covered with paper and/or absorbent pads. If the animal can be encouraged to urinate/defecate when in this “run” it will help reduce contamination of the cage and facilitate easier clean up of urine/feces once the dog is returned to the primary cage.

#### SUPPLIES TO BE BROUGHT INTO HOT ZONE AS NEEDED (UP TO TWICE DAILY)

	Item	Quantity	Freq
	Animal food	1 meal	BID
	Animal water	Refill water jug (~4 L)	PRN
	Biohazard bags	1-2	PRN
	Absorbent pads	1-2	SID-BID
	<b>Replenish any supplies used from warm and hot inventories noted above</b>		PRN

#### SUPPLIES TO BE BROUGHT INTO HOT ZONE FOR SAMPLE COLLECTION

	Item	Quantity
	Needles (2 sizes)	2 of each
	Syringes (appropriate size(s))	2 of each
	Small sharps container	1
	Small bottle or isopropyl alcohol	1
	Blood collection tubes (plastic)	2 of each needed
	Fecal cups	2
	Urine cups	2
	Syringe to collect urine from floor	1

All items to remain in hot zone once brought in, except for sample tubes & containers themselves.



Additional receptacles (bag or container) to be left in warm zone and cold zone for exiting samples from hot zone and warm zone, respectively.

### Appendix 3: List of disinfectants and their efficacy against Ebola virus

Disinfectant	Concentration	Contact Time	Comments
Accelerated hydrogen peroxide	As per label.	5 minutes	Not tested against Ebola virus but effective against more hardy non-enveloped viruses, safe and relatively stable in organic debris.
Bleach	1:10 – 1:50 dilution of household (5.25% bleach)	≥ 10 minutes	Highly effective disinfectant when used properly. Inactivated by organic debris and light. Corrosive and potentially noxious in small spaces.
Quaternary ammonium disinfectant	Product-specific	Product-specific. Often 30 minutes	Highly variable group of disinfectants. Only consider products with demonstrated efficacy against non-enveloped viruses.

## Appendix 4: Procedures for donning personal protective equipment (PPE)

### Donning PPE

1. **Remove Personal Clothing and Items:** Change into surgical scrubs (or a similar garment such as coveralls) and dedicated washable (plastic or rubber) footwear in a clean area. All personal items (e.g., jewelry, watches, cell phones, pagers, pens) should be removed.
2. **Engage Trained Observer:** A trained observer will be present to confirm visually that PPE is appropriate, intact and has been donned properly. A checklist can facilitate this process.
3. **Ensure all PPE is present:** In conjunction with the observer, ensure that all required PPE items, and items required for the doffing process, are present.
4. **Inspect PPE Prior to Donning:** Visually inspect the PPE to ensure that it is intact and is appropriate condition.
5. **Perform Hand Hygiene:** Perform hand hygiene with an alcohol-based hand sanitizer (AHS). For each use of AHS in donning and doffing, allow hands to dry before moving to the next step.
6. **Put on Inner Gloves:** Put on first pair of gloves.
7. **Put on hooded suit:** Ensure the suit is large enough to allow unrestricted freedom of movement. Ensure cuffs of inner gloves are tucked under the sleeve of the suit. Tape the zipper, leaving a long tab, to facilitate suit removal.
8. **Put on Boot Covers**
9. **Secure suit to boot covers with duct tape.**
10. **Put on Outer Gloves:** Put on second pair of gloves (with extended cuffs). Ensure the cuffs are pulled over the sleeves of the suit.
11. **Secure suit to outer gloves with duct tape**
12. **Put on N95 Respirator or equivalent respiratory protection:** Fit testing must have been performed on the specific respirator that is being used. Perform seal test.
13. **Put on Outer Apron (if used):** An outer apron may be considered if there is an increased risk of contact with the animal's body fluids.
14. **Put on Face Shield and/or Goggles:**
15. **Verify:** The trained observer and user should verify that all PPE is correctly placed. The user should also verify that they can move in an unrestricted manner, including extending the arms and bending at the waist.

1.

## Appendix 5: PPE doffing

### Doffing PPE

1. **Prior to exiting the hot zone**, apply AHS or an appropriate disinfectant to the outer gloves and provide the indicated contact time. Spray the bottom of the boot covers with disinfectant. Wipe any areas of gross contamination with disinfectant.
2. **Engage Trained Observer**: The trained observer will confirm that PPE is intact, identify areas of gross contamination and confirm that the doffing process is correct. The observer should read aloud each step in the doffing process. A written checklist can facilitate this process.
3. **Inspect**: Inspect the PPE for visible contamination, cuts, or tears. If any PPE is visibly contaminated, apply disinfectant and provide the indicated contact time before proceeding.
4. **Disinfect Outer Gloves**: Disinfect outer-gloved hands with either an appropriate disinfectant or AHS. Provide the required contact time for the disinfectant or allow the alcohol to dry before proceeding.
5. **Remove Apron (if used)**: Remove and discard apron taking care to avoid contaminating gloves by rolling the apron from inside to outside. Inspect PPE under the apron for damage or visible contamination. If contamination is noted, disinfect as per step 2.
6. **Disinfect Outer Gloves**: Disinfect outer-gloved hands with either a disinfectant or AHS.
7. **Loosen duct tape on boot covers and outer gloves.**
8. **Remove Suit**: While outer gloves and boot covers are still attached to the suit, remove the suit carefully by rolling the suit outward and downward. Avoid contact of garments worn underneath the suit with the outer surface of suit during removal. Touch only the inside of the suit. Remove the suit, outer gloves and boot covers as one unit. Place all removed PPE in a biohazard bag that is placed in a rigid, impermeable container with a lid.
9. **Inspect and Disinfect Inner Gloves**: Inspect the inner gloves' outer surfaces for visible contamination, cuts, or tears. *If an inner glove is visibly soiled, cut, or torn*, then disinfect the glove with either an appropriate disinfectant wipe or AHS. Then remove the inner gloves, perform hand hygiene with AHS and don a clean pair of gloves. *If no visible contamination, cuts, or tears are identified* on the inner gloves, then disinfect the inner-gloved hands (leaving gloves on the hands) with either an appropriate disinfectant wipe or AHS.
10. **Remove Face Shield and/or Goggles**: Remove the full face shield or goggles by tilting the head slightly forward, grabbing the rear strap and pulling it over the head, gently allowing the face shield/goggles to fall forward and discard. Avoid touching the front surface of the face shield/goggles.
11. **Disinfect and Change Inner Gloves**: Disinfect inner gloves with either an appropriate disinfectant wipe or AHS. Remove and discard gloves taking care not to contaminate bare

hands during removal process. Perform hand hygiene with AHS. Don a new pair of inner gloves.

12. **Remove Respirator:** Remove the respirator by tilting the head slightly forward, grasping first the bottom tie or elastic strap, then the top tie or elastic strap, and remove without touching the front of the respirator. Discard disposable respirators. If a re-usable respirator is used, it should be disinfected as per manufacturer recommendations.
13. **Disinfect and Remove Inner Gloves:** Disinfect inner-gloved hands with either an appropriate disinfectant wipe or AHS. Remove and discard gloves taking care not to contaminate bare hands during removal process.
14. **Perform Hand Hygiene:** Perform hand hygiene with AHS.
15. **Inspect:** The trained observer should inspect the person for any evidence of contamination of the surgical scrubs or coveralls. If contamination is identified, the area should be sprayed with disinfectant.
16. **Scrubs:** Scrubs should be removed and placed directly into a biohazard bag or similarly identified, impermeable bag for autoclaving
17. **Shower:** A shower is recommended to complete the process.

## Appendix 6: References and suggested resources

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## Resources

Barton Behravesh et al, Interim guidance for dog or cat quarantine after exposure to a human with confirmed Ebola virus disease. <http://www.cdc.gov/vhf/ebola/pdf/dog-cat-quarantine.pdf>

Barton Behravesh et al, Interim guidance for public health officials on pets of Ebola virus disease contacts. <http://www.cdc.gov/vhf/ebola/pdf/pets-of-ebola-contacts.pdf>

Centers for Disease Control and Prevention, Interim guidance for environmental infection control in hospitals for Ebola virus. <http://www.cdc.gov/vhf/ebola/hcp/environmental-infection-control-in-hospitals.html>

Centers for Disease Control and Prevention, Interim guidance for specimen collection, transport, testing and submission for persons under investigation for Ebola virus disease in the United States.

<http://www.cdc.gov/vhf/ebola/hcp/interim-guidance-specimen-collection-submission-patients-suspected-infection-ebola.html>

Centers for Disease Control and Prevention. Guidance on personal protective equipment to be used by healthcare workers during management of patients with Ebola virus disease in US hospitals, including procedures for putting on (donning) and removing (doffing). <http://www.cdc.gov/vhf/ebola/hcp/procedures-for-ppe.html>

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